

Supporting information for:

Harnessing Monocytes for Liposomal Rosiglitazone-Mediated Anti-Inflammatory Effect

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- Figure S1 Formulation attempts using the passive loading method
- Figure S2 Optimization of drug encapsulation using the active loading method
- Figure S3 Internalization of LipRhod-Cy5 in RAW264.7 cells
- Figure S4 Dose-response initial experiments

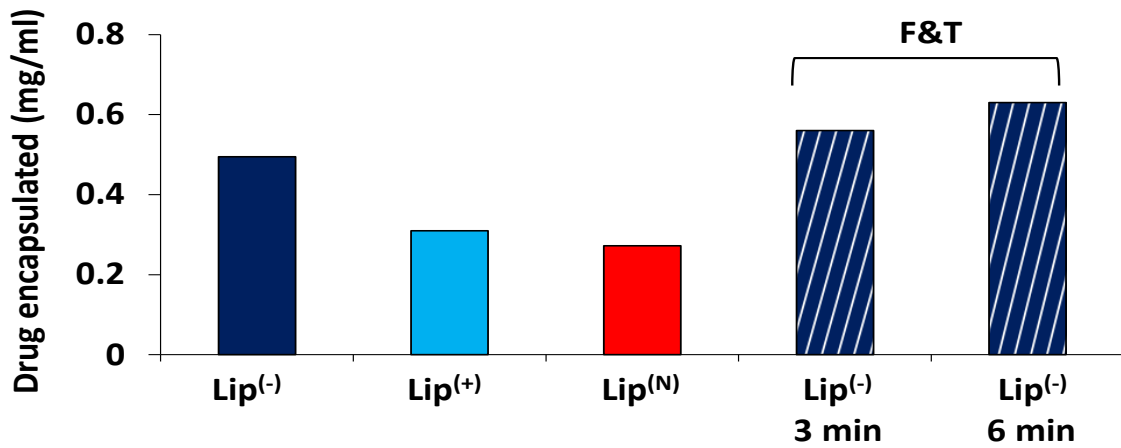


Figure S1: Formulation attempts by the passive loading method and additional step of the freeze and thaw technique (F&T) for preparing liposomes containing rosiglitazone; Lip(-), DSPC:DSPG:cholesterol (negatively charged); Lip(+), DSPC:DOTAP:cholesterol (positively charged); Lip(N), DSPC:DPPC:cholesterol (neutrally charged); 3 and 6 min refers to the freezing and thawing time, respectively.

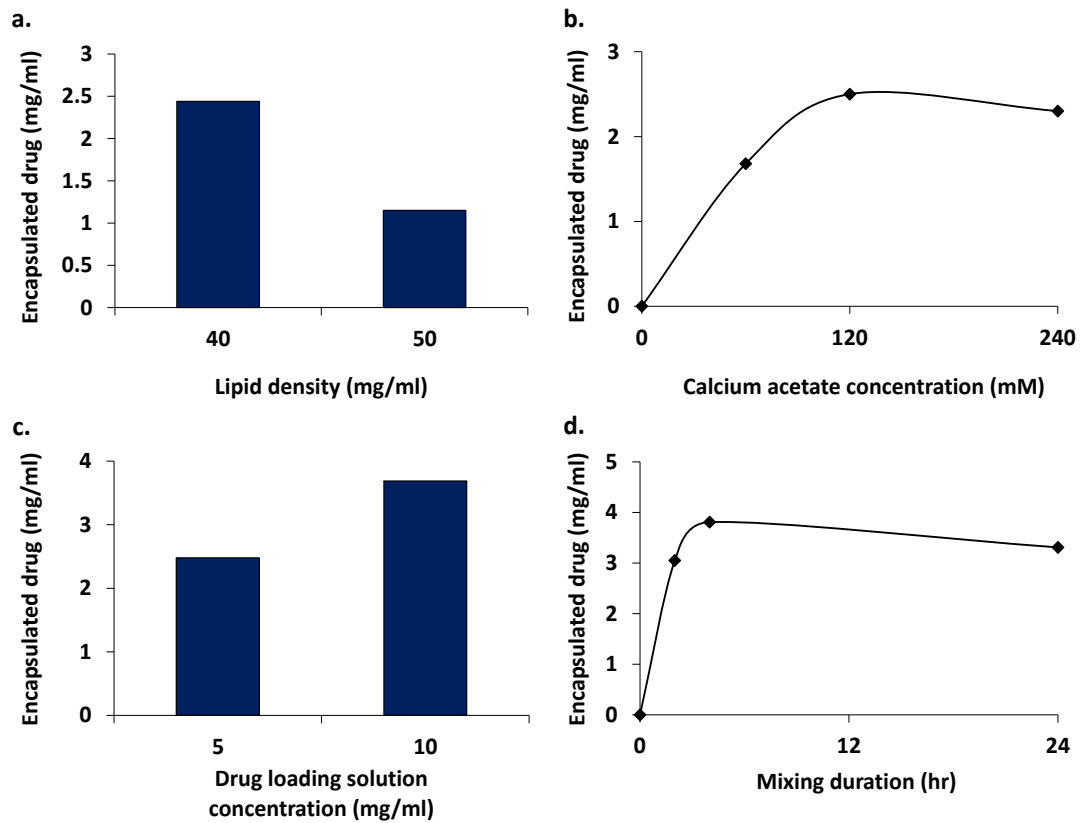


Figure S2: Optimization of rosiglitazone encapsulation using the active loading method. (a) Effect of lipid density; (b) Effect of hydrating calcium acetate concentration; (c) Effect of drug loading concentration; (d) Effect of mixing duration.

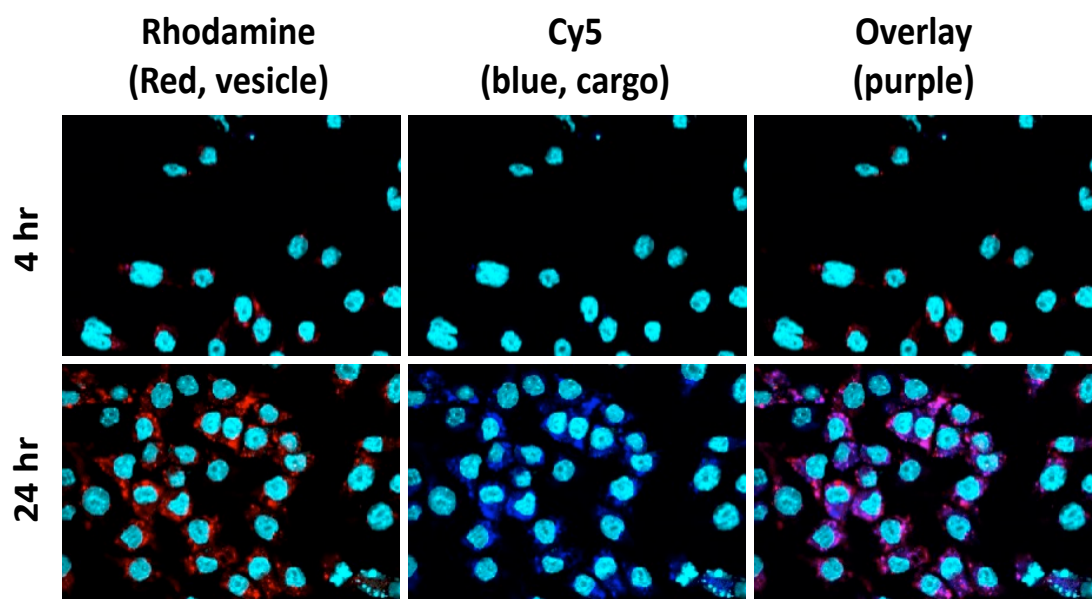


Figure S3: Qualitative assessment of fluorescently-labeled empty liposomes (rhodamine and Cy5, aqueous core and membrane, respectively; LipRhod-Cy5; 0.5mg/ml lipids concentration) uptake by RAW264.7 cells. The nuclei is shown in turquoise (Hoechst), liposomes bilayer is shown in red (PE-Rhodamine), liposomes cargo is shown in blue (Cy5), and overlay of vesicle and cargo is shown in purple. Images were obtained by means of confocal microscopy and analyzed with FV10-ASW 3.1 viewer software. The fluorescence intensity is normalized to non-treated

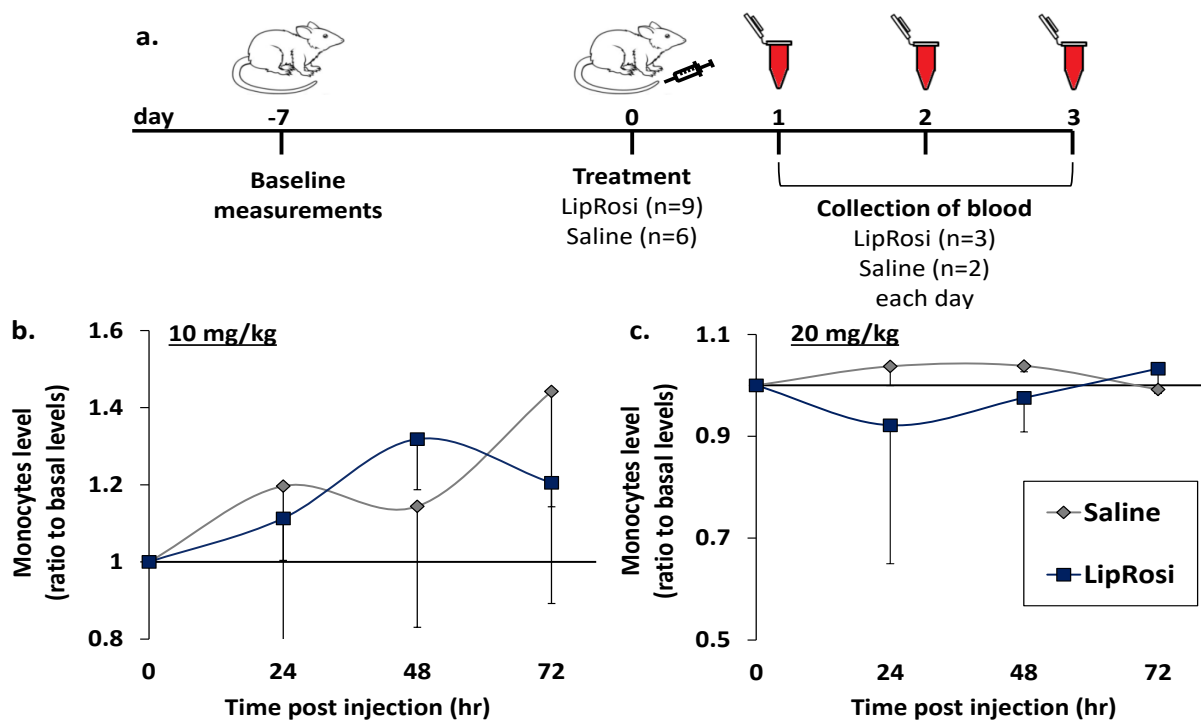


Figure S4: The effect of Liposomal rosiglitazone on circulating monocytes level in intact rats at a dose of 10 mg/kg (**b**) and 20 mg/kg (**c**). (**a**) Schematic presentation of animal experimental setup. (**b,c**) Monocytes levels measured in the blood a week before injection (baseline, 0 hr), 24, 48 and 72 hr post single injection (IV) by means of flow cytometry. Monocytes were defined as ED1-positive (CD68) cells and are presented as the ratio normalized to the individual baseline level. Data is presented as the mean \pm SEM. n=5 animals in each group.